PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference VA/H-33271A	FOR FURTHER ACTION	See Form PCT/IPEA/416
International application No. PCT/EP2004/008464	International filing date *(day/month/year)* 28.07.2004	Priority date *(day/month/year)* 29.07.2003
International Patent Classification (IPC) or national classification and IPC C07K14/285, C12N15/31		
Applicant NOVARTIS AG et al.		

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of 8 sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
   a. ☐ *sent to the applicant and to the International Bureau)* a total of sheets, as follows:
      ☐ sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
      ☐ sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.
   b. ☐ *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in computer readable form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).

4. This report contains indications relating to the following items:

- ☒ Box No. I Basis of the opinion
- ☐ Box No. II Priority
- ☐ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☒ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- ☐ Box No. VI Certain documents cited
- ☐ Box No. VII Certain defects in the international application
- ☐ Box No. VIII Certain observations on the international application

Date of submission of the demand	Date of completion of this report
18.02.2005	25.01.2006
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized Officer Donath, C Telephone No. +49 89 2399-8710

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/EP2004/008464

Box No. I Basis of the report

1. With regard to the **language**, this report is based on the international application in the language in which it was filed, unless otherwise indicated under this item.

   ☐ This report is based on translations from the original language into the following language , which is the language of a translation furnished for the purposes of:
   - ☐ international search (under Rules 12.3 and 23.1(b))
   - ☐ publication of the international application (under Rule 12.4)
   - ☐ international preliminary examination (under Rules 55.2 and/or 55.3)

2. With regard to the **elements*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

Description, Pages

1-22 as originally filed

Claims, Numbers

1-25 as originally filed

Drawings, Figures

1, 2 as originally filed

☒ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing

3. ☐ The amendments have resulted in the cancellation of:
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

* *If item 4 applies, some or all of these sheets may be marked "superseded."*

Box No. IV Lack of unity of invention

1. ☐ In response to the invitation to restrict or pay additional fees, the applicant has:
   - ☐ restricted the claims.
   - ☐ paid additional fees.
   - ☐ paid additional fees under protest.
   - ☐ neither restricted nor paid additional fees.
2. ☐ This Authority found that the requirement of unity of invention is not complied with and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.
3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is
   - ☐ complied with.
   - ☒ not complied with for the following reasons:

     see separate sheet
4. Consequently, this report has been established in respect of the following parts of the international application:
   - ☒ all parts.
   - ☐ the parts relating to claims Nos. .

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	3,4,11-13,16,22,24,25
	No:	Claims	1,2,5-10,14,15,17-21,23
Inventive step (IS)	Yes:	Claims	
	No:	Claims	1-25
Industrial applicability (IA)	Yes:	Claims	1-25
	No:	Claims	

2. Citations and explanations (Rule 70.7):

   see separate sheet

Supplemental Box relating to Sequence Listing

Continuation of Box I, item 2:

1. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application and necessary to the claimed invention, this report has been established on the basis of:

   a. type of material:

   ☒ a sequence listing

   ☐ table(s) related to the sequence listing

   b. format of material:

   ☒ in written format

   ☒ in computer readable form

   c. time of filing/furnishing:

   ☐ contained in the international application as filed

   ☐ filed together with the international application in computer readable form

   ☒ furnished subsequently to this Authority for the purposes of search and/or examination

   ☒ received by this Authority as an amendment on15.11.2004

2. ☒ In addition, in the case that more than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional observations, if necessary:

Ad section IV.:

The international preliminary examining authority is of the opinion that the application does not comply with the requirements of unity as set forth in the PCT regulations (Article 34(3), Rule 13 PCT). It will be considered that the following separate alleged inventions or groups of inventions are not so linked as to form a single general inventive concept:

1) claims 1-14,24 and 25 refer to an isolated or purified 55 kDa extracellular protein of *Photobacterium damselae* subsp. *piscicida* having apoptogenic properties, its amino acid sequence, the nucleic acid sequence encoding the same, DNA expression vector comprising said nucleic acid sequence, a vaccine comprising either an immunogenic derivative of said protein or the above DNA expression vector, antibodies raised against said protein, and the use of either the amino acid sequence or the nucleic acid sequence of the protein for the manufacture of a test for diagnosis of infection with *Photobacterium damselae* subsp. *piscicida* or of pasteurellosis in fish.

2) claims 15-23 refer to a general method of preparing a vaccine against pasteurellosis comprising a step of growing *Photobacterium damselae* subsp. *piscicida* cells in culture; further the claims refer to a vaccine composition comprising an inactivated cell culture supernatant or extracellular protein preparation rich in p55 from *Photobacterium damselae* subsp. *piscicida*.

The general inventive concept underlying the two above mentioned inventions of the present international application can be seen as the reference to *Photobacterium damselae* subsp. *piscicida* and to the identification of a 55 kDa extracellular protein within said bacterium.
However, this general inventive concept is not novel having regard to the state of the art as illustrated by document WO-A-01/10459 which discloses a vaccine comprising an extracellular 55 kDa protein from *Photobacterium damselae* subsp. *piscicida* for the prophylactic and/or therapeutic treatment of fish for infection by the organism *Photobacterium damselae* subsp. *piscicida* (see WO-A-01/10459, p.1,l.3-8; p.4, l.7 - p.7, l.35; p.16, l.7 - p.17, l.24; Fig.3 and 7).

Therefore, a single general inventive concept is not acceptable, making necessary to reconsider the technical relationship or interaction between the different inventions mentioned.
This leads to their regrouping under different subjects as listed above, each subject is falling under its own inventive concept, being a solution to the problem in a way which differs from the state of the art.

Ad section V.:

1. The following documents are cited:

   D1 WO-A-01/10459
   D2 EP-A-0 773 295
   D3 WO-A-96/12734
   D4 Aquaculture 120(3-4), 201-208, 1994
   D5 J.Appl.Ichthyol. 14(3/4), 265-268, 1998

2. First, the present International application refers to an isolated or purified 55 kDa extracellular protein of *Photobacterium damselae* subsp. *piscicida* having apoptogenic properties, its amino acid sequence, the nucleic acid sequence encoding the same, DNA expression vector comprising said nucleic acid sequence, a vaccine comprising either an immunogenic derivative of said protein or the above DNA expression vector, antibodies raised against said protein, and the use of either the amino acid sequence or the nucleic acid sequence of the protein for the manufacture of a test for diagnosis of infection with *Photobacterium damselae* subsp. *piscicida* or of pasteurellosis in fish.
   Second, the present International application refers to a general method of preparing a vaccine against pasteurellosis comprising a step of growing *Photobacterium damselae* subsp. *piscicida* cells in culture; further the claims refer to a vaccine composition comprising an inactivated cell culture supernatant or extracellular protein preparation rich in p55 from *Photobacterium damselae* subsp. *piscicida*.
   In view of the documents cited in the International Search Report only the subject-matter of claims 3,4,11-13,16,22,24 and 25 of the present International application

has to be regarded as being new (Article 33(2) PCT).

2.1 D1 discloses a vaccine comprising an extracellular 55 kDa protein from *Photobacterium damselae* subsp. *piscicida* for the prophylactic and/or therapeutic treatment of fish for infection by the organism *Photobacterium damselae* subsp. *piscicida*. Also antibodies were raised against the 55 kDa extracellular protein (see WO-A-01/10459, p.1,l.3-8; p.4, l.7 - p.7, l.35; p.16, l.7 - p.17, l.24; Fig.3 and 7). Although no sequence data are available for said 55 kDa protein described in D1 the applicant is informed that the amino acid sequence of a protein is considered only as a parameter which does not render the protein as such novel over the prior art. Even if the applicant will provide prove that the 55 kDa protein of D1 is different from the one comprising the amino acid sequence as shown in SEQ ID NO:2 the protein of D1 has to be regarded as an immunogenic derivative thereof.

Thus, the above document is novelty-destroying for the subject-matter of claims 1,2,5-10,14,15,17-19 and 23.

2.2 D3 describes the preparation of a vaccine against pasteurellosis starting from the bacterium *Pasteurella piscicida*. Different vaccine preparations have been compared to each other, those wherein the cells have been cultured without iron supplementation and in the absence of iron chelating agents, and those wherein the cells have been cultured in medium with added $FeCl_3$ or with an added iron chelator agent (see D3, p.2, l.33 - p.5, l.11; p.6, l.5 - p.7, l.5; claims 1,4,5,9,17,20).

Thus, the above document is novelty-destroying for the subject-matter of claims 15,17-21 and 23.

2.3 D4 discloses a comparative study of the efficacy of two vaccine formulations, a whole-cell bacterin and a toxoid-enriched whole-cell vaccine against *Pasteurella piscicida*. By this study the role of the extracellular products (ECP) as protective antigens against this pathogen has be evaluated. In order to prepare the respective vaccine formulations the cells were cultured in normal medium without iron supplementation and in the absence of iron chelating agents and the ECP were inactivated with formaldehyde (see D4, p.202-204,'Materials and methods').

Thus, the above document is novelty-destroying for the subject-matter of claims 15,17-21 and 23.

3.1 The closest prior art to evaluate the inventiveness of claims 3,4,11-13,24 and 25 is the above cited document D1.

The subject-matter of these claims only differs from the teachings of D1 in that it refers to the nucleic acid sequence encoding the 55 kDa extracellular protein and to the use of either the amino acid sequence, the nucleic acid sequence or the antibodies raised against said protein for the manufacture of a test for diagnosis of infection with *Photobacterium damselae* subsp. *piscicida* or of pasteurellosis in fish. However, D2 discloses immunization of cultured fish by DNA expression systems. DNA plasmids containing sequences encoding antigenic components, such as sequences encoding en extracellular protein of *Pasteurellosis* are introduced by transfection into aquaculture species. Also mentioned are pharmaceutical compositions comprising DNA vaccines in an amount effective for the treatment and prevention of diseases caused by pathogens such as *Pasteurella piscicida* (see D2, p.3, l.45 - p.6, l.51).
Thus, in view of the teachings of D1 in combination with that of D2 the subject-matter of claims 3,4,11-13,24 and 25 is considered to lack an inventive step.

3.2 The closest prior art to evaluate the inventiveness of claims 16 and 22 is either the above cited document D3 or D4.

The subject-matter of these claims only differs from the teachings of D3 or D4 in that it refers to a vaccine preparation wherein the used cells are grown until mid-exponential phase instead of being grown until late-exponential growth phase.
D5, however, discloses that in order to prepare vaccine formulations from *Pasteurella piscicida* the cells can be collected form various phases of the exponential growth (see D5, p.266, 'Experimental trials').
Thus, in view of the teachings of D3 or D4 in combination with that of D5 the subject-matter of claims 16 and 22 is considered to lack an inventive step.